SIGMA CHEMICAL COMPANY

Product No. A-1043

Lot 106H4857

Anti-Chicken IgG (whole molecule)
Alkaline Phosphatase Conjugate
Antibody developed in Rabbit
IgG Fraction of Antiserum

Antiserum is developed in rabbit using IgG isolated from pooled normal chicken serum as the immunogen. The antibody is isolated from rabbit anti-chicken IgG antiserum by immunospecific purification to remove essentially all rabbit serum proteins, including immunoglobulins, which do not specifically bind to chicken IgG. Rabbit anti-chicken IgG is conjugated to Sigma Alkaline Phosphatase using 0.2% glutaraldehyde. The conjugate is provided as a solution in 0.05 M Tris buffer, pH 8.0, containing 1% BSA, and 1 mM $MgCl_2$, with 0.1% sodium azide (see MSDS)* as preservative.

Specificity

Specificity of the anti-chicken IgG antibodies for chicken IgG is determined by immunoelectrophoresis (IEP) prior to conjugation using normal chicken serum and chicken IgG.

Identity and Purity

Identity and purity of the antibody is established by immunoelectrophoresis, prior to conjugation. Electrophoresis of the antibody preparation followed by diffusion versus anti-rabbit IgG and anti-rabbit whole serum result in single arcs of precipitation in the gamma region.

Titers

1. 1:15,000 (Direct ELISA)

Titer is defined as the dilution of conjugate sufficient to give a change in absorbance of 1.0 at 405 nm after 30 minutes of substrate conversion at 25°C (Voller, et al.[1]). Microtiter plates are coated with purified chicken IgG at a concentration of 5 µg/ml in 0.01 M phosphate buffered saline, pH 7.4, containing 0.1% sodium azide.

Substrate: *p*-Nitrophenyl phosphate (pNPP, Sigma Product No. N-2765), 1.0 mg/ml in 10% diethanolamine buffer, pH 9.8, containing 0.01% $MgCl_2$ and 0.02% NaN_3.

2. Dot Blot

a. A dilution of 1:60,000 was determined by direct assay using 40 ng chicken IgG/dot.
b. A dilution of 1:80,000 was determined by indirect assay using 20 ng human IgG/ dot and chicken anti-human IgG as the primary antibody.
c. In an indirect chemiluminescence system using 10 ng human IgG/dot and chicken anti-human IgG as the primary antibody, this product was determined to have a dilution of 1:80,000 when used as secondary antibody. 1,2-Dioxetane and enhancer was used as substrate.

3. Immunohistology

A dilution of 1:20 was determined by indirect assay on formalin-fixed, paraffin-embedded human tonsil sections using chicken anti-human IgG as the primary antibody.

Working Dilution

Working dilution should be determined by titration assay. Due to product improvement and changes in the assay procedure, we now list a lot specific titer by direct ELISA for this product. Due to differences in assay systems, this titer may not reflect the user's actual working dilution.

Storage

Store at 2-8°C. **Do Not Freeze**.

Reference

1. Voller, A., et al., Bulletin WHO, **53**, 55 (1976).

*Due to the sodium azide content a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazards and safe handling practices.